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Recent diagnostic challenges in schistosomiasis

Současné výzvy v diagnostice schistosomóz

Bachelor's thesis

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Authorship statement

I declare that I am the only independent author of the present bachelor's thesis and that I referenced all used information sources and literature accordingly. I further declare that the present thesis has not been submitted anywhere else in order to obtain an academic title.

Prague, 29.05.2020

Prohlášení

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V Praze, 29.05.2020 Jana Liberdová

ABSTRACT

Schistosomiasis is debilitating waterborne helminthiasis of global medical and veterinary importance. It is caused by digenetic trematodes of the genus *Schistosoma*. More than 250 million people are currently infected, and 700 million people are at daily risk of acquiring this neglected tropical disease. Schistosomiasis is prevalent mostly in sub-Saharan Africa, particularly in poor communities living in impoverished conditions with limited access to safe water. The global control of this disease relies on single drug-oriented preventive chemotherapy campaigns. Changing epidemiological situation together with the general nature of this parasitosis (long-term development in several phases, multiorgan pathogenesis with complex manifestations) makes its diagnosis difficult. Current diagnostic standards may be unreliable in many specific settings, and their universal use needs to be reconsidered. In schistosomiasis diagnostics, the choice of specific methodology should be made accordingly to the particular situation requirements. The present thesis reviews recent approaches and challenges in schistosomiasis diagnostics.

Key words: schistosomiasis, diagnostics, neglected diseases, global control, elimination

ABSTRAKT

Schistosomóza je závažné onemocnění celosvětového lékařského a veterinárního významu. Tuto nemoc způsobují dvou hostitelské motolice rodu *Schistosoma*. Více než 250 miliónů lidí je v současnosti nakaženo a 700 milionů lidí tato zanedbávaná tropická nemoc denně ohrožuje. Schistosomóza je rozšířená hlavně v subsaharské Africe, a to zejména u chudých obyvatel žijících ve špatných podmínkách s omezeným přístupem k čisté vodě. Celosvětová kontrola této nemoci je zařizována hlavně pomocí preventivních terapeutických kampaní jednoho léku. Měnící se epidemiologická situace společně s obecnou povahou této parazitózy (dlouhodobý vývoj s několika fázemi, multiorgánová patogeneze s komplikovanými projevy) činí její diagnostiku problematickou. Současné diagnostické standardy mohou být ve spoustě situací nespolehlivé a jejich univerzální využití by se mělo přehodnotit. Pro diagnostiku schistosomóz platí, že výběr konkrétní metodiky by se měl provádět na základě nároků jednotlivých diagnostických situací. Tato práce shrnuje současné přístupy a výzvy v diagnostice schistosomóz.

Klíčová slova: schistosomóza, diagnostika, opomíjené nemoci, celosvětová kontrola, eliminace

Contents

INTRODUCTION

Water and infectious diseases, the two more or less connected factors play an essential role in our everyday life as well as in global economy. Limited access to safe water and poor sanitation is a fatal combination. Millions of people suffer from water-related morbidities (WHO, 2008). Although significant progress against infectious diseases has been made, the death rates and burdens, especially in developing countries, remain high (Ritchie and Roser, 2019). The current global situation regarding both of these aspects including water pollution, extensive draught, along with world pandemics urge renewed attention. Anyone, regardless of social or economic status, can be affected if water shortage happens in their environment and can become infected if they come in contact with waterborne disease.

One of the most important waterborne diseases, which clearly connects both introduced factors is schistosomiasis. Studies dedicated to schistosomiasis are valuable not only for dealing with this severe condition, but also to alert issues in global water management. Diagnosis, as an initial step in coping with infectious diseases, remains challenging in schistosomiasis. The choice of test should be ideally based on patients' signs and symptoms, other medical conditions, and history, which may be difficult to determine during schistosomiasis infection. In practice, this suggestion is often not followed. There are many different tests available, some to mention are excreta and tissue biopsy examinations, blood tests including serology, molecular diagnostics, and medical imaging. A combination of these tests may be required to confirm the diagnosis (CDC, 2018).

The aim of the present thesis is to (1) introduce the current situation of schistosomiasis disease, (2) present available and novel diagnostic approaches with emphasis on the advantages and drawbacks of these methods in various settings, (3) provide an up-to-date review of recent diagnostic challenges.

1. SCHISTOSOMIASIS

Schistosomiasis is a debilitating infectious parasitic disease of global medical and veterinary importance. This helminthiasis belongs to the group of neglected tropical diseases even though it is the second major parasitic infection after malaria (Chitsulo et al., 2004).

Schistosomiasis has been affecting humans since ancient times. One of the oldest evidence of schistosomiasis is reported from northern Syria and goes back more than 6000 years ago. The infection occurrence was indicated in Egyptian medical papyri from around 1500 years BC as well as in the reports from ancient Mesopotamia (Di Bella et al., 2018). In modern history, German professor Theodor Billharz first identified infective parasites causing this condition in Cairo in 1851. In his honor, the disease is historically referred to as bilharziasis or bilharzia (McManus et al., 2018; Tan and Ahana, 2007). In the 1980s, efficient antischistosomal chemotherapy was introduced and subsequently enhanced therapeutic approaches and control initiatives.

Despite the decades of research advances and elimination efforts, schistosomiasis remains a severe threat to global health with alarming morbidity and mortality. Persistent prevalence and high virulence of this parasitosis represent a significant burden for many, especially developing countries (Ross et al., 2002; Chitsulo et al., 2000).

1.1. General information

Schistosomiasis is a waterborne disease caused by parasitic trematode flatworms of the genus *Schistosoma*. Six schistosome species are largely responsible for human infections. *Schistosoma haematobium*, *S. mansoni*, *S. japonicum* are three the most common species and *S. mekongi*, *S. intercalatum*. *S. guineensis* are common only focally (Colley et al., 2014). Less importantly, other schistosomes such as *Trichobilharzia* spp. with avian definitive hosts can incidentally infect humans and provoke skin allergic reactions. However, in humans as noncompatible host, they are unable to continue the life cycle and cause chronic disease (Kolářová et al., 2013).

Schistosome parasites have a complex digenetic life cycle, including multiple larval stages and adult sexual dimorphism (see figure 1).

Besides remarkable adult worm pairs, well known from the books, schistosomes appear in five main different developmental stages – egg, miracidium, sporocyst, cercaria, and schistosomula (Ross and Yuesheng, 2016). Adult worms, inside the definitive host, have a tubular body 10 to 20 mm long with an oral and ventral sucker, an outer tegument, and a blind-ending digestive system. The schistosomes sexes can be easily differentiated. Male worms are short, muscular, and thick, with robust suckers. Female worms are slenderer and more elongated. In the mature state, male worms clasp females in their gynecophoral canal, creating together the mating *copula*. Without this pairing, the female worm is unable to develop, and lay eggs (Ross and Yuesheng, 2016; Lewis and Tucker, 2014; Becker et al., 1980).

The eggs laid by female adult worms are round to oval in shape, in size approximately 180 x 70 µm, and contain evolving nascent miracidia. Morphological variations of the eggs can be used to differentiate the individual *Schistosoma* species (WHO, 2019a; Ross et al., 2002). Miracidia hatched from the eggs in the freshwater environment are oval, motile, ciliated larvae approximately 200 µm long. Sporocysts are found in the intermediate snail host body and appear like growing pleomorphic sack bodies that form immature evolving cercariae. Mature cercariae are the second motile larvae of the freshwater environment, approximately 500 µm long with a body part containing penetration glands, and a potent forked tail. Schistosomulae are adaptive juveniles inside the definitive host tissues with a worm-like body, initially approximately 100 µm long that undergo massive morphological changes and maturation, resulting in the development of the adult worm pairs able to reproduce sexually (Ross and Yuesheng, 2016; Lewis and Tucker, 2014).

Humans are considered to be a predominant mammalian host for *S. mansoni*, *S. intercalatum* and *S. guineensis*. However, these schistosomes can also infect other mammals, particularly rodents (Standley et al., 2012). It is presumed that *S. haematobium* develops exclusively in humans and possibly other primates. In contrast, *S. japonicum* and *S. mekongi* have a zoonotic cycle with a broad spectrum of definitive hosts, such as dogs, cattle, livestock and other domestic animals, making the management of the disease much more difficult (Catalano et al., 2018; Standley et al., 2012). These parasites develop through specific intermediate snail host of the genera *Biomphalaria* for *S. mansoni*; *Bulinus* for *S. haematobium*, *S. intercalatum*, *S.guineensis*; *Oncomelania* for *S. japonicum*; and *Neotricula* for *S. mekongi* (CDC, 2019; Chitsulo et al., 2004). However, these findings need more investigation as the host specificity could vary during the evolution of the species. Understanding the schistosome lifecycle and relationships between its hosts is fundamental in moving towards the elimination of this disease (CDC, 2019; Standley et al., 2012; Gryseels et al., 2006).

Initial infection occurs if the humans are exposed to the water containing the cercariae. Infectious cercariae penetrate the skin. Within hours after contact with contaminated water, maculopapular lesions known as cercarial dermatitis may develop. This skin rash usually disappears spontaneously after a few hours to days (Ross et al., 2007; Lambertucci et al., 1997). Further exposures to the same species may appear with a stronger allergic reaction (Kolářová et al., 2013; Chamot et al. 1998). In the body, cercariae develop into the juvenile schistosomulae. Schistosomulae leave the skin through blood and lymphatic vessels subsequently carried by the venous circulation to the lungs. Subadult worms exit the lungs after several days and enter the hepatic portal system, where they finish the maturation and mating, and continue the downstream migration to the target location of the blood system depending on the certain *Schistosoma* species preference (CDC, 2019; Ross and Yuesheng, 2016; Lewis and Tucker, 2014).

Schistosoma haematobium is responsible for urogenital schistosomiasis, and the adult worms are most often located in the vesicular and pelvic venous plexus of urogenital system. The rest of the human schistosomes cause intestinal schistosomiasis, with adult worms settled in the mesenteric venules. *S. mansoni* worms are often located in the lower mesentery close to the large intestine, as well as *S. intercalatum* and *S. guineensis*, which rather occurs in the lowest parts. *Schistosoma japonicum* prefers the upper mesentery close to the small intestine (CDC, 2019). While inside the patient, the adult worms can reproduce and lay eggs up to 40 years (Colley et al., 2014).

Schistosome eggs are either excreted into the environment with feces or urine (usually during the initial stages of the infection) or they become trapped in host tissues (usually during the chronic stages of the infection). From the eggs reaching freshwater, miracidia hatch and may infect a susceptible intermediate freshwater snail host, in which they complete asexual replication. After 4 to 6 weeks, tens of thousands of cercariae are released into the water from the snails, and the life cycle continues (CDC, 2019). On the other hand, in the chronic phase, the eggs retained in the host tissues do not leave the body and provoke a strong host's immune responses (Schwartz and Fallon, 2018; Ross et al., 2002).

First systemic manifestations of the infection are known as acute schistosomiasis so-called Katayama syndrome, usually occurring 14 to 84 days after exposure (Ross et al., 2007). This condition is more common in non-immune travelers experiencing a primary infection than in immunized residents of endemic areas, who could remain asymptomatic (Bottieau et al., 2006; Gryssels et al., 2006; Ross et al., 2002). It may correspond with the parasites' maturation and migration inside the patient's body, subsequently provoking a hypersensitive reaction to their antigens. The symptoms are usually flu-like (Jauréguiberry et al., 2010).

Over time, untreated schistosomiasis proceeds to its chronic phase. The symptoms at this stage correlate with the load and location of the highly immunogenic eggs trapped in the body tissues. So, egg retention and granuloma formation during chronic schistosomiasis can cause multi-organ pathologies such as fibrosis (McManus et al., 2018; Ross et al., 2002). Later stages of fibrotic and collagen accumulation are more severe than the initial inflammatory syndrome and may not be quite reversible (Schwartz and Fallon, 2018). Additional clinical syndromes can be caused by ectopic egg deposition (Gryseels et al., 2006).

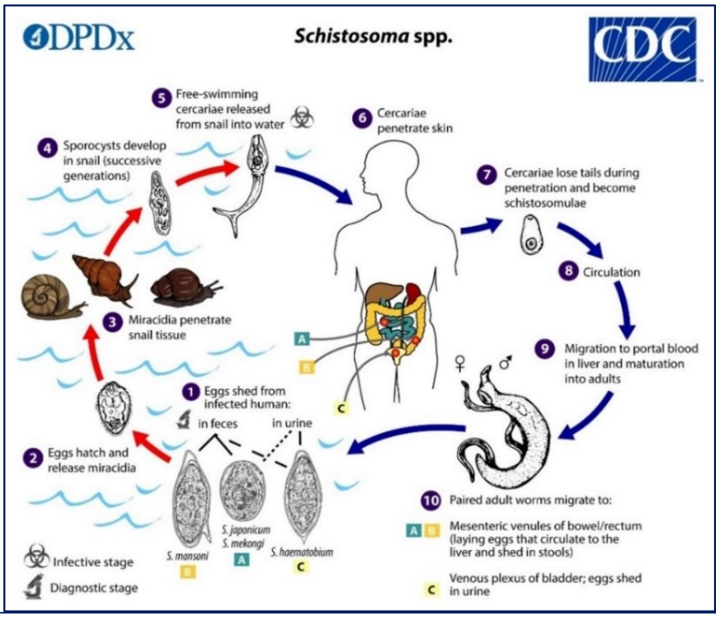

Figure 1 / *Schistosoma* spp. life cycle

(1) Eggs laid by female adult schistosomes are excreted from humans in either feces or urine. (2) From the eggs reaching freshwater environment, larvae miracidia hatch. (3) Miracidia actively swim and infect the susceptible intermediate snail hosts. (4) In the intermediate hosts, miracidia evolve through two generations of sporocyst into the (5) cercariae larvae, which are released back to the freshwater environment. (6) Infective cercariae actively search and penetrate the skin of definitive hosts. (7) After the penetration, cercariae evolve into juvenile schistosomulae. (8) Schistosomulae then migrate from the skin through the body's circulation system to the lungs and heart, (9) afterward mature into the adult worms in the liver and continue through the portal vein system to the final location. (10) Paired adults reside and copulate in the mesenteric veins. The exact location depends on the certain species preference (A) *S. japonicum*, (B) *S.mansoni*, (C) *S. haematobium*. Figure and information adapted from CDC, 2019.

1.2. Epidemiology

As mentioned before, schistosomiasis is one of the most prevalent human parasitic infections with significant morbidity and mortality (King, 2009). Over **250 million** people worldwide are currently infected, and nearly triple that number, over **700 million** people are at risk of acquiring the infection (Steinmann et al., 2006). Chronic schistosomiasis is the most prevalent form, with more than **20 million** people suffering from severe symptoms (Chitsulo et al., 2000). The estimated global burden of this disease is **3,31 million** disability-adjusted life years (DALYs*). According to WHO, yearly schistosomiasis mortality only in African countries could be as high as **280 000 deaths** (WHO, 2010; King and Bertino, 2008). But, for example, the Global Burden of Disease (GBD) data estimates only **11 000 deaths** per year, showing a great underestimation of this issue in some studies (Molyneux et al., 2017).

WHO reports 78 countries with currently ongoing transmission and estimates that approximately 90 % of people in need of anti-schistosomal therapy live in Africa (WHO, 2020a). Intestinal disease caused by *S. mansoni* occurs in most African countries, the Middle East, the Caribbean, and South American countries such as Brazil, Venezuela, and Surinam. Other intestinal schistosomes are distributed more locally. *Schistosoma intercalatum* and *S. guineensis* are found within the central African rainforests. *Schistosoma japonicum* is prevalent in the Western Pacific region (China, Indonesia, the Philippines), and *S. mekongi* is endemic along the Mekong River basin in Southeast Asia (Cambodia, Laos). Urinary disease caused by *S. haematobium* occurs in most African countries and the Middle East (WHO, 2020a; Engels et al., 2002; Chitsulo et al., 2000), autochthonous cases were also reported from Corsica (Noël et al., 2018). Schistosomiasis has been eradicated from Japan (Tanaka and Tsuji, 1997), and has been eliminated to very low transmission levels in the Lesser Antilles, Tunisia, Morocco, and Turkey (WHO, 2019b). Interventions made in China (Wang et al., 2009) and Egypt (Barakat, 2013) also show progress in fighting the disease. Up-to-date situation and trends can be tracked online at the Global Health Observatory website (WHO, 2019b).

As the previous paragraph indicates, due to the ecological requirements of schistosomes, endemic areas are limited to tropical and subtropical zones. Schistosomiasis is characterized by heterogeneous prevalence and focal epidemiology (Fenwick and Jourdan, 2016; Gryseels et al., 2006). The highest transmission rates persist within populations living in poor conditions, with limited access to safe water (King, 2010). However, the risk of acquiring infection is known from luxurious tourist resorts as well (Corachan, 2002). Such a case was reported from Lake Malawi shores, where many individuals, especially travelers, became infected (Cetron et al., 1996). Freshwater lakes and rivers are considered as a major source of the disease, but also manmade water systems and agricultural practices (i.e., work at rice fields) represent substantial transmission sites (Steinmann et al., 2006). Subpopulations of residents with common daily water-related habits such as bathing, washing, laundry, fishing, etc. keep high disease prevalence and reinfection rates (Chandiwana and Woolhouse, 1991), up to 100 % in some communities (Bakuza et al., 2017).

Schistosomiasis demography varies with age. The highest prevalence and infection intensities are among children and young adolescents of endemic areas. The average age of the first infection in endemic regions is 2 years of age. As the parasites then colonize the patient's body, the worm burden

*DALYs = Disability Adjusted Life Years is quantifying metrics used in measuring disease burden and health conditions. The number of DALYs represents the number of "healthy" life years lost due to disability, morbidity and mortality (WHO, 2020b).

increases approximately for the next 10 years (Colley et al., 2014). For example, in Sudan, the highest infection rates were reported in children aged 10 to 13 years old (Hajissa et al., 2018). In further adulthood, there is a general decline of both prevalence and infection intensity, but chronic symptoms may persist. It might correlate with a stronger immune system response (Mazigo et al., 2012) or age-related changes in water exposure (Bajiro el at., 2016). Among travelers, the most infected ones are usually young adults (Lingscheid et al., 2017), which likely relates to the practiced adventurous type of traveling (Röser et al., 2018). In terms of gender demography, schistosomiasis is more prevalent in males of endemic countries, probably because of more frequent water exposure during fishing or agricultural activities (Mazigo et al., 2012). However, for example, genital lesions caused by *S. haematobium* are more life-threatening for women than men (Nour, 2010). Furthermore, congenital infection from mother to child has also been reported (Friedman et al., 2007).

1.3. General challenges

Several issues in schistosomiasis remain to be solved.

It is supposed that metrics currently used to describe epidemiological situation do not reflect the actual cost of this human infection (King and Bertino, 2008). Due to missing accurate data from many areas, global prevalence and infection risk estimates are made based on extrapolations (Engels et al., 2002). Also, the official mortality of schistosomiasis is underestimated, as mentioned above in the chapter (1.2).

In addition to direct morbidities, schistosomiasis often occurs with related comorbidities (Colley et al., 2014). Concurrent infection with schistosomes can affect physiological and pathological relations and immunological response between the host and co-infecting pathogens (Abruzzi and Fried, 2011). Some studies suggest a protective effect of schistosomiasis infection, for example, against severe malaria attacks (Nmorsi et al., 2009; Briand et al., 2005). However, in general, subsequent infections have increased severity (Courtin et al., 2011; Midzi et al., 2010; Muok et al., 2009). Schistosomiasis is associated with HIV and HPV infections (Mosunjac et al., 2003), viral hepatitis (Gasim et al., 2015), and malaria (Wilson and Dunne, 2012). Furthermore, schistosome worms are considered as potent carcinogens, particularly in squamous cell carcinoma and hepatocellular carcinoma (van Tong et al., 2017). Subtle morbidities related to schistosomiasis such as chronic anemia, malnutrition, infertility, growth and development impairment also represent a significant burden for infected communities (Hotez et al., 2009; King and Dangerfield-Cha, 2008). Although better schistosomiasis control comes along with better management of all mentioned associated complications, these issues often remain neglected and separated (Wilson and Dunne, 2012; King and Dangerfield-Cha, 2008).

Also, the spread and reemergence of schistosomiasis are currently broadly discussed themes. Increasing population and its movement together with water-resource and agricultural engineering, are the main reasons for transmission growth and disease introduction to new locations in endemic countries (Chitsulo et al., 2000). Intense migration and ongoing environmental changes create a risk of schistosomiasis establishment at new sites in non-endemic countries (Stensgaard et al., 2019; Adeel, 2017). It is presumed that these global trends will escalate the spread of infectious diseases worldwide (Kim et al., 2014). Expansion of tropical and sub-tropical disease vectors to higher latitudes, including Europe, is already happening (ECDC, 2012). Snail vectors, due to their limited dispersal ability highly dependent on the ecological factors, have been overlooked so far. Plus, most of the human contacts with freshwater in developed countries are restricted to recreation or agriculture, and water treatment plus sanitation are generally provided with a high standard. Therefore, the spread of schistosomiasis might come unexpected (Kincaid-Smith et al., 2017). There is a well-known reported case of schistosomiasis emerge northward, to Corsica (France). During the last outbreak there in the summer of 2013, more than 100 cases were confirmed (Noël et al., 2018). Furthermore, the assumption of non-human reservoirs along with possible hybridization of zoonotic *S. bovis* with the anthropogenic *S. haematobium* and their adaptation to the new environments represent a threat of disease expansion (Oleaga et al., 2019).

Knowledgeable of schistosomiasis epidemiology from the chapter (1.2), we can classify it as a poverty-related disease. The most vulnerable populations live in impoverished conditions with limited safe water, sanitation, hygiene, and healthcare provision (Grimes et al., 2015; King, 2010). Only a small improvement in these basic settings could help significantly with the interruption of transmission and prevention of schistosomiasis (Campbell et al., 2018). Unfortunately, in a lot of countries it is still not happening. Recurrent and multiple pathogen infections are everyday standard for the people of highly endemic countries, representing an enormous burden for the national healthcare and economy (King, 2010). Schistosomiasis affects people's life from an early age. Its morbidities are linked to impaired education abilities and poor physical performance leading to reduced individual productivity (Hotez et al., 2009; King and Dangerfield-Cha, 2008). Chronically infected communities have to sacrifice their entire capital to the bare survival; anything above that is unaffordable. Therefore, the overall population prosperity is minimal (Stothard et al., 2013; King and Dangerfield-Cha, 2008). Break out of this cycle, especially in sub-Saharan African countries, seems impossible (Adenowo et al., 2015; Engels et al., 2002). For the illustration, the prevalence of *S. haematobium* in Nigerian households with an income above 1600 USD is only 1,5 %, but in those with an income below 600 USD, it is alarming 70 % (WHO, 2015). All of these poverty-related issues create another problem. Affected populations cannot pay for their treatment, which limits commercial incitement to invest in product development for diseases of poverty (Weber et al., 2019). Pharmaceutical and biotechnology businesses are mostly focused on more profitable markets, then neglected tropical diseases (Pedrique et al., 2013). In many areas, the only available one-sided interventions focused on morbidity are alone not enough to fight this disease (King, 2010).

Infected patients can be treated with Praziquantel (PZQ). This quinoline-pyrazine derivate is an anti-schistosomal drug of choice since its introduction in the 1970s (Gryseels et al., 2006). It is currently characterized by its exclusive and extensive use in mass drug administration (MDA) campaigns, which are an essential part of global schistosomiasis control (Bergquist et al., 2017). The success of PZQ results

from its many positive features. It is an effective, verified, easy-to-use, affordable pill without any harsh side effects. Praziquantel has low toxicity and is safe for young children and pregnant women. No long-term difficulties were observed in patients. Praziquantel acts against the adult worms of all *Schistosoma* spp (Cioli et al., 2014; Colley et al., 2014; Cioli and Pica-Mattocia, 2003). However, it is not a flawless pill. Praziquantel is ineffective against immature schistosomes, non-prophylactic, only short-term effective, and cannot prevent reinfection. Therefore, the outcomes of MDA campaigns are often not optimal (Tebeje et al., 2016; Cioli et al., 2014). If targeted patients are hosting schistosome larvae during the treatment or if they come in contact with the cercariae later again, the disease is not treated nor interrupted (Cioli et al., 2014; Cioli and Pica-Mattocia, 2003). Generally recommended standard PZQ dose is 40 to 60 mg/kg. Optimization of this regimen for pediatric use is missing and should be considered as the highest treatment failures are reported among children (Stothard et al., 2013).
Schistosomiasis control programs rely on this single drug treatment for nearly 40 years now. Intensive PZQ use among millions of people repeatedly brings up a threat of drug resistance, which is necessary to avoid, a variety of approaches would be welcome (Bergquist et al., 2017). Praziquantel-resistant schistosomes are currently known mostly from the laboratory settings (Cupit and Cunningham, 2015; Lotfy et al., 2015; Doenhoff et al., 2002), although the appearance of PZQ-tolerant strains recovered from infected patients with poor treatment response was also reported (Doenhoff et al., 2002; Ismail et al., 1999). Another resistance issue that rises with schistosomiasis control is the use of common treatment combination of PZQ and antimalarial drug Artemisinin, which is active against immature schistosomes. This treatment regimen could potentially induce the artemisinin resistance in *Plasmodium* spp. (Colley et al., 2014). All these therapy-related issues highlight the need to continue the research and development of new reliable anthelmintic therapies for all different purposes (Rollinson et al., 2013).

Another major challenge is the development and optimization of diagnostic methods for schistosomiasis, which I would like to review in more detail at the following pages.

2. DIAGNOSTIC METHODS

Accurate diagnostics is the first step in coping with the diseases. It is also an essential part of their management and control (McCarthy et al., 2012). Moreover, improved and advanced diagnostic methods are important not only in clinical diagnoses but also in research and product development (Colley et al., 2014).

Generally, schistosomiasis is diagnosed by microscopic detection of eggs in stool and urine samples. This direct method is commonly performed along with antibody detection, especially in non-endemic areas (WHO, 2020a; CDC, 2019). Schistosomiasis can also be diagnosed by parasite antigen and DNA detection (Weerakoon et al., 2015). However, these tests require further optimization to be widely implemented (Bergquist et al., 2016; Utzinger et al., 2015). In specific conditions, additional diagnostic techniques can help with infection confirmation (McManus et al., 2018).

Besides other important values of diagnostic methods reviewed in this text, sensitivity and specificity are often highlighted. Briefly, sensitivity represents the test capacity to correctly diagnose schistosomiasis positive patients, and specificity represents the test capacity to correctly exclude schistosomiasis negative patients (Weerakoon et al., 2015).

See figure 2, which summarizes laboratory-based diagnostic tests for schistosomiasis and some of their characteristics described in the following chapters (2.1 to 2.4) and provides a general overview in the table for clarity.

Test	Sample	Quantitative	Sensitivity	Specificity	Target settings	Cost
Detection of eggs by light microscopy						
Kato-Katz	fresh stool	yes	low	high	endemic	moderate
FECT	fresh stool	no	moderate	high	endemic	moderate
MHT	fresh stool	yes	low/moderate	high	endemic	low
Urine filtration	fresh urine	yes	low/moderate	high	endemic	low
Detection of specific antibodies						
ELISA	serum	no	high	moderate	non-endemic	moderate
IHT	serum	no	high	moderate	endemic	moderate
Detection of specific antigens						
ELISA[a]	serum, urine, vaginal lavage	yes	high	high	non-endemic	high
POC-CCA	serum, urine	semi[b]	high	high	endemic	low
UCP-LF CAA	various clinical samples	yes	high	high	endemic[c]	moderate
Detection of parasite DNA						
PCR	various clinical samples	yes	high	high	non-endemic	high
LAMP	stool, urine	yes	high	high	endemic[c]	high

Figure 2 / Summary of reviewed laboratory-based diagnostic tests for *Schistosoma* spp. infection

[a]For CCA and CAA antigens. [b]Semi-quantitaive trace results of 1+, 2+, 3+ could be interpreted from this rapid test. [c]Further optimization required.

2.1 Detection of eggs by light microscopy

Microscopic detection of schistosome eggs in feces for intestinal schistosomiasis and urine for urogenital schistosomiasis, or additionally in tissue biopsies for chronic stages of both is the diagnostics of choice for an established schistosomiasis infection (McManus et al., 2018).

The eggs are relatively easy to detect. As mentioned before in chapter (1.1), it is possible to determine individual species based on the eggs' morphological characteristics (WHO, 2019a; Ross et al., 2002). Although microscopic detection of the eggs is a direct and highly specific approach, the main drawback remains its low sensitivity. In light infections, the numbers of eggs are often too low to be detectable (McManus et al., 2018; Bärenbold et al., 2017). False negatives then result in an underestimation of the disease prevalence and inaccurate MDAs progress assessment (Bergquist et al., 2009). Eggs shedding occur after approximately 4 to 8 weeks post-infection and varies markedly during as well as between the days (Bärenbold et al., 2017; Bergquist et al., 2016). The results of microscopic detection change with the number of specimens examined. For instance, in one study, sensitivity increased from 57 % for one specimen up to 95 % for four specimens (Bärenbold et al., 2017).

Schistosomiasis infection should not be excluded by the excreta examinations alone (Weerakoon et al., 2015; Colley et al., 2014).

The WHO-recommended approach for **stool examinations** in endemic countries is the Kato-Katz method (see figure 3) based on microscopic detection of eggs in sieved and stained standardized stool samples (WHO, 2019a; WHO, 2013a). In brief, a small amount of feces is pressed through the nylon screen, and this sieved fecal material is collected with a spatula. In the next step, the collected feces are filled into template hole on a microscopic slide, the template is then removed, and remaining stool cylinder is covered with a glycerol soaked cellophane piece and firmly pressed to spread evenly. After approximately 20 minutes, glycerol clears the feces, what is necessary to check. A clarified clean sample is the one through which newspaper print is possible to read. Such a prepared sample is systematically examined under the microscope, detected eggs are determined and counted (WHO, 2019a).

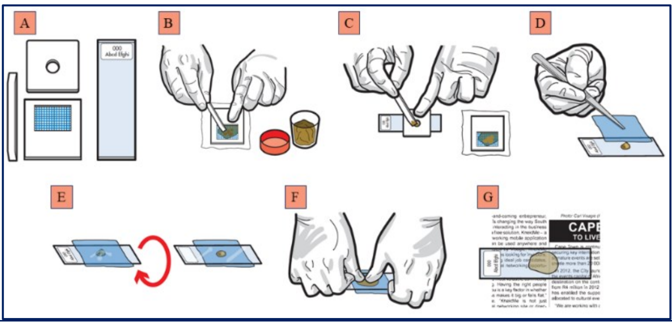

Figure 3 / Kato-Katz technique format provided by WHO for laboratory and field workers

(A) Plastic spatula, stainless steel/plastic/cardboard template with a hole, stainless steel/nylon/plastic screen with mesh, microscope slide. (B) Press the fecal material through the screen, scrape the sieved material with a spatula. (C) Place the template in the center of the labeled microscope slide, fill the hole with the sieved fecal material. (D) Remove the template, place the glycerol soaked cellophane piece over the fecal sample. (E) Invert the microscope slide. (F) Firmly press on a smooth, hard surface to spread the feces. (G) Pick up the slide and place it with the cellophane strip upwards. The sample is clear if the newspaper print is possible to read through it. Figure and information adapted from WHO, 2019a.

Kato-Katz thick fecal smear technique is a widely used method for diagnosing intestinal schistosomiasis and also other soil-transmitted helminthiases (STHs; Ross et al., 2002). The number of detected eggs is used to calculate the "eggs per gram" (EPG) value to classify the infection intensity (McManus et al., 2018). According to WHO, more than 400 EPG found in the smear means heavy infection, 400 to 101 EPG moderate infection, and under 100 EPG light infection (WHO, 1993). This field-friendly technique with high specificity and relatively low cost is recommended for diagnosis and evaluation of schistosomiasis in endemic areas (WHO, 2019a; Gray et al., 2011). However, its application is limited. As already mentioned, lacking sensitivity, especially in light infections, is currently the main issue as many patients are treated regularly during MDAs, reducing the infection intensities. Kato-Katz technique is also highly dependent on staff training (Bergquist et al., 2009). Improving the sensitivity with several smears examinations is time-consuming and increases the cost, therefore not affordable in limited settings (Ajibola et al., 2018; Rollinson et al., 2013).

Another way to improve the sensitivity of microscopic methods is to add concentration procedures, which increase the chance to detect light infections. These techniques help to separate the eggs from the fecal matter, making them easier to detect (Weerakoon et al., 2015). Formalin-ether concentration technique (FECT) is often used in intestinal schistosomiasis diagnostics. This method's main steps include the homogenization of feces, filtration, sieving, and centrifugation. Such a prepared sample is ready for microscopic analysis (Utzinger et al., 2015). Although concentration techniques are very useful, they increase the cost, preparation time, and laboratory equipment requirements of the diagnostics (Ajibola et al., 2018).

An additional approach for detecting eggs in stool samples is the miracidia hatching test (MHT), which could be beneficial in settings with no suitable microscopic equipment. This specialized hatching technique is based on the strong phototropic behavior of the miracidia. In this procedure, a fecal sample or homogenate of tissue suggested with the eggs' presence is diluted in water. Flask with diluted material is placed in the dark, with only an upper collection site illuminated where the hatched miracidia will assemble. Miracidia obtained by this method could be observed with a simple stereomicroscope, magnifying glass, or eventually by an experienced eye. Other positive features are the low cost and the possibility to use also diarrheic feces. (Nelwan, 2019; Jurberg et al., 2008). However, hatching techniques are relatively time-consuming and influenced by external conditions such as temperature and water quality (Bergquist et al., 2016). MHT is established and employed mainly in China (Ross et al, 2002).

The WHO-recommended approaches for **urine examinations** are sedimentation and filtration. A simple technique to confirm urogenital schistosomiasis is the microscopic detection of eggs in a sedimented or centrifugated urine sample (WHO, 2019a; Gryseels et al., 2006). Yet, urine filtration is considered the gold standard. During this technique, well shaken (10 mL) of urine is squeezed out from a syringe on a polycarbonate filter with a defined pore size. Eggs retained on the filter can be stained with Lugol's iodine. Orange-stained eggs are determined and counted under the microscope (WHO, 2019a; Utzinger et al., 2015). Similarly to the Kato-Katz, the infection intensity is classified by the number of eggs per 10 mL of urine. More than 50 eggs detected in the sample indicate heavy infections, and less than 50 light infections (WHO, 1993). Another similarity with the thick smear techniques is test simplicity, but lack of sensitivity, especially in light infections (Ajibola et al., 2018; Weerakoon et al., 2015). A recent study reports successfully performed ultra-low-cost urine filtration using readily available paper products such as paper towels and newspaper instead of more expensive laboratory filters, showing great potential for the application in limited-resource settings (Ephraim et al., 2014).

Histopathological studies of affected **tissue biopsies** (e.g., liver, urinary bladder) are important in schistosomiasis diagnostics, particularly at the chronic when the egg excretion decreases and tissue-trapped egg pathologies prevail (Gray et al., 2011; Ross et al., 2002). These microscopic observations contribute to the diagnosis confirmation and also to the understanding of the disease progress and pathology (McManus et al., 2018).

2.2 Detection of specific antibodies

Antibody-based assays proved their importance in clinical diagnosis, particularly in travel medicine (Colley et al., 2014). They are also useful for control in low-endemicity areas close to the interruption of transmission, and for monitoring of the disease re-emergence (Bergquist et al., 2009; Ross et al., 2002). Optimally used antibody-based techniques are highly sensitive (Le and Hsieh, 2017; Gryssels et al., 2006). Anti-schistosomal antibodies can be detected in patients with no present egg excretion, such as those with early or light infections (Le and Hsieh, 2017; Ross et al., 2002). The production of specific antibodies begins approximately 4 to 7 weeks post-infection (McManus et al., 2018). Antibody levels in the patients change only slowly and can persist years after successful treatment (Gryssels et al., 2006). Therefore, serology is unable to distinguish between present and past infections and can cross-react with other helminthiases (Le and Hsieh, 2017; Rollinson et al., 2013). The specificity varies with the type of test and the antigen selection which is not standardized, unified and often differs among the laboratories and suppliers (Utzinger et al., 2015; Jauréguiberry et al., 2010).

The selection of antigens for antibody-based testing is still a matter of research. As schistosomes in the patient's body mature and develop, slightly different antigens provoke the immune system's reactions in each phase of infection. Consequently, diverse antigens from any developmental stage should be used in serological assays (Hinz et al., 2017; Le and Hsieh, 2017). Egg and adult worm antigens are standardly used in commercially available testing kits (Gray et al., 2011). Larval (cercarial, schistosomulum) antigens are useful in diagnosing early infections, particularly cercarial transformation fluid (CTF) shows high sensitivity when used as an antibody target. Antibody detection based on circulating schistosome antigens also has promising results (Hinze et al., 2017; Bergquist et al., 2016). Various recombinant, synthetic and non-schistosome antigens are being studied to achieve the highest test accuracy (Hinz et al., 2017).

One of the well-established serologic tests for detecting proteins is enzyme-linked immunosorbent assay (ELISA). This colorimetric test is available in many modifications for various applications (Utzinger et al., 2015). Anti-schistosomal ELISA kits are designed to determine the presence of antibodies against *Schistosoma* spp. antigens in serum. In brief, the ELISA procedure starts by coating microtiter plate wells with selected antigens (Hinz et al., 2017). If there are anti-schistosomal antibodies present in the tested sample, they will create antigen-antibody complexes, which could be visualized by using the correct enzyme conjugate and its substrate resulting in the color-changing catalytic reaction. This reaction is then stopped by adding an acidic solution. Final color density is analyzed and correlates with the presence of antibodies (Bergquist et al., 2016). Considering its complex protocol and equipment requirements, ELISA is more suitable for research and clinical use rather than field application (Weerakoon et al., 2015).

Another antibody-based assay used in schistosomiasis diagnostic is the indirect hemagglutination test (IHT). In the IHT testing kits, the erythrocytes sensitized with selected schistosomal antigens agglutinate in the presence of serum anti-schistosomal antibodies, creating distinctly visible coating in the microplate well (Weerakoon et al., 2015). Due to its relative simplicity, IHT is a favorable antibody-based test for field community screenings (Hinz et al., 2017; Bergquist et al., 2016).

Other assays detecting antibodies are to mention, immunofluorescent assay (IFA) during which the immunocomplexes consisting of antibodies tagged with a fluorescent dye and targeted antigens are visualized by a fluorescent microscope, and circumoval precipitin test (COPT) which is based on observation of patient serum precipitation around schistosomal eggs (Hinz et al., 2017; Weerakoon et al., 2015). There is a wide choice of antibody-based approaches with some similarities and differences available for schistosomiasis diagnostics. In conclusion, these tests' results depend greatly on the design, methodology, target antibody class, antigen selection, and the basic rules of immunological response. A well-balanced assay can show many aspects of the infection (Hinz et al., 2017; Utzinger et al., 2015; Cavalcanti et al., 2012).

2.3 Detection of specific antigens

Detection of circulating schistosomal antigens is a promising direct approach that may eventually replace current diagnostic standards in some applications (Gray et al., 2011; Ross et al., 2002). Field adjusted, point-of-care (POC*) antigen-based tests are valuable tools for epidemiological situation monitoring, control campaigns targeting and evaluation (Weerakoon et al., 2015; Rollinson et al., 2013). Antigen-based testing can be performed with variety of samples such as serum, urine, or sputum. These methods are both highly specific and sensitive. The levels of serum antigens correlate with the living worms burden, infection intensity, plus they decrease quickly after the successful treatment reflecting the therapeutic response (Bergquist et al., 2016; Weerakoon et al., 2015). The main flaw of these methods remains their relatively high cost and need for further standardization and commercialization to be used routinely (Le and Hsieh, 2017; Cavalcanti et al., 2012).

Commonly detected antigens in schistosomiasis diagnostics are adult worm antigens (AWA), soluble egg antigens (SEA), and circulating antigens (Weerakoon et al., 2015). Circulating anodic (CAA) and circulating cathodic (CCA) antigens are well determined in established infections and show reliable results in diagnostic applications (McManus et al., 2018; Bergquist et al., 2016).

Besides antibody detection, ELISA can also be used to detect specific antigens. The basic principle of this approach is the same as described above (chapter 2.2). However, for antigen detection, the sandwich ELISA is usually used. The main change in the procedure is that the microplate wells are coated with antibodies to detect the wanted antigen, antigen-antibody complexes are then visualized by adding secondary antibody-enzyme conjugate (Hinz et al., 2017). Antigen-based ELISA is a valuable

*POC = Point-of-care testing means it is available right at the sample collection site, with results ready within a few minutes rather than hours (Bergquist et al., 2016).

research tool. As already mentioned, due to its complex requirements, it is not very suitable for field applications (Bergquist et al., 2016; Weerakoon et al., 2015).

There is currently commercially available rapid CCA-based assay in the convenient form of dipstick or cassette ready for POC testing (Le and Hsieh, 2017). The principle of POC-CCA is the detection of antigen presented in a urine or serum sample by an immobilized labeled antibody. After approximately 20 minutes of incubation, the resulting immunocomplexes create a visible ready-to-read line on a testing membrane (Bergquist et al., 2016). This approach is especially promising in diagnosing intestinal schistosomiasis from urine samples. With further adjustments and validation, POC-CCA can be beneficial in prevalence mapping, community screening, and control program evaluations in *S. mansoni* endemic and co-endemic regions (McManus et al., 2018; Le and Hsieh, 2017; Colley et al., 2014).

Another relatively convenient antigen-based test is an up-converting phosphor lateral flow (UCP-LF CAA) assay. The principle of UCP-LF CAA is the detection of antigen presented in the clinical sample by UCP-antibody reporter conjugates immobilized on a nitrocellulose membrane. After incubation, the results can be analyzed using a portable reader (Bergquist et al., 2016; Utzinger et al., 2015). The possibility of using a dry-reagent format makes this approach more practical for distribution and storage (Utzinger et al., 2015). Well-optimized UCP-LF CAA can be valuable for testing in low-endemicity areas close to the elimination (Le and Hsieh, 2017).

As the antigen-based diagnostic approaches show promising results, they are in the current research focus. Diverse antigens are being selected, and novel assays are being designed and tested (Hinz et al., 2017; Weerakoon et al., 2015).

2.4 Detection of parasite DNA

DNA detection represents one of the most sensitive and specific diagnostic approaches known. DNA-based testing has a capacity to provide clinical diagnoses in all phases of schistosomiasis infection as well as to monitor the epidemiological situation and evaluate the control programs (Nelwan, 2019; Ajibola et al., 2018). Various clinical samples (e.g., body fluids, excreta) can be used for DNA-based testing (Le and Hsieh, 2017; Gray et al., 2011). Multiplex DNA analysis of concurrent parasitoses is a valuable method for diagnosing patients with numerous infections (McManus et al., 2018). The DNA detection is now increasingly employed in high-resource settings. However, the introduction and optimization of these methods for limited-resource settings in endemic countries might represent one of the biggest challenges to overcome (McManus et al., 2018; Le and Hsieh, 2017).

The primary method for detecting DNA is polymerase chain reaction (PCR). This approach is extensively used with many designs and application variations, including highly accurate diagnostics of infectious diseases. In short, the principle of PCR is the amplification of targeted specific DNA fragment during the cyclic procedure of denaturation, hybridization, and elongation of the DNA strand by DNA polymerase (Weerakoon et al., 2018; Bergquist et al., 2016). Schistosome DNA can be detected by PCR in alternative clinical samples (vaginal lavage, semen, rectal snip, etc.), making this approach advantageous in diagnosing ectopic schistosomiasis (Le and Hsieh, 2017; Utzinger et al., 2015). This method allows early diagnosis from about one week post-infection, evaluation of the therapeutic response, as well as the distinction of individual schistosome species, strains, and hybrids (Weerakoon et al., 2018; Weerakoon et al., 2015; Utzinger et al., 2015). Although PCR is still an out-of-reach method for large-scale field use, research to bring PCR out of the laboratory environment seems promising (Weerakoon et al., 2018; Rollinson et al., 2013).

Another appealing DNA-based method for schistosomiasis diagnostics is loop-mediated isothermal amplification (LAMP). This method is carried out as a rapid single-tube test under isothermal conditions that results in DNA amplification (Weerakoon et al., 2018). In contrast with PCR, LAMP has reduced equipment requirements, and the results are ready to read visually (Le and Hsieh, 2017). Similarly to PCR, LAMP also has the capacity to detect the infection from early phases and evaluate treatment response (Weerakoon et al., 2015). This approach has the potential to become a molecular diagnostic of choice for field use (Le and Hsieh, 2017). However, further optimization and validation in community-based testing are necessary (Weerakoon et al., 2018).

Research, development, and improvement of DNA-based diagnostics come with other positive side-effects. Schistosomes' genomic data is now more available and can be used in other studies, for example, to identify vaccine and therapy targets, to inspect drug-resistance, or simply to see the molecular background of any chosen characteristic (Weber et al., 2019; Weerakoon et al., 2018).

2.5 Other diagnostic approaches

Besides laboratory-based techniques, schistosomiasis questionnaires are useful for rapid mapping, control programs targeting and evaluation in highly endemic areas (Rollinson et al., 2013; Bergquist et al., 2009). A few simple questions survey (e.g., Did you experience bloody urine within the last month? yes/no) to self-report the infection is an accessible and reliable approach with satisfying results. Note to say, questionnaires can be simultaneously used as educational material for the interviewed patients (Le and Hsieh, 2017).

Valuable rapid tool for advanced field diagnostics in endemic areas are urine reagent dipsticks, designed to detect hematuria and proteinuria (albumin, creatinine) in the urine sample (Weerakoon et al., 2015). This approach shows great results in community-based testing for *S. haematobium* infections (Le and Hsieh, 2017; Utzinger et al., 2015). A combination of questionnaires and urine reagent dipsticks is very successful, especially in diagnosing urogenital schistosomiasis due to the strong correlation between the infection and hematuria (Utzinger et al., 2015; Weerakoon et al., 2015). These methods together make a good tool for prevalence mapping and identification of transmission hot spots (Le and Hsieh, 2017).

On the other side of the diagnostics spectrum, there are medical imaging and radiology studies that represent sensitive and specific approaches for clinical evaluation of disease-associated pathology.

Techniques such as endoscopy, ultrasonography (US), magnetic resonance imaging (MRI), computed tomography (CT), echocardiography (ECHO), etc. are used to visualize pathological changes associated with chronic schistosomiasis as well as ectopic schistosomiasis (Gray et al., 2011; McManus et al., 2018). These pathologies are to mention pulmonary infiltrates, liver fibrosis (Symmer's fibrosis), urogenital tract lesions and carcinoma, tissue edema, among others. Imaging-based methods are not only useful in visualizing the pathologies but also in therapy evaluation, prediction of risks and complications, and consideration of eventual surgical procedure (McManus et al., 2018; Gryssels et al., 2006). Disposition and cost of these techniques limit their use in resource-restricted healthcare facilities of endemic countries. However, mobile units and practical guidelines have been developed and proved very useful in field diagnostics (WHO, 2000; Norseth et al., 2014).

Complete blood count (CBC) is a method used in hospitals for clinical diagnosis. This test may reveal some of the schistosomiasis indicators, such as eosinophilia, particularly in acute syndrome (Gray et al., 2011; Ross et al., 2007). Furthermore, anemia, thrombocytopenia, prolonged prothrombin time, hyperglobulinemia, hypoalbuminemia, higher serum urea and electrolytes are prominent in severe chronic stages of the disease (Gray et al., 2011; Ross et al., 2002).

In specialized hospital laboratories and travel clinics, cytokine spectrum as a schistosomiasis disease characteristic can be used for diagnostics. In acute stages of the infection, proinflammatory cytokines such as TNF-alpha, IFN-gamma, IL-1, IL-2 could be measured by ELISA, later during the chronic end-stages IL-4, IL-5, IL-10, IL-13 are dominant. However, the cytokine response switch is not general. This approach is considered as additional for clinical diagnosis, yet is widely used in research settings (McManus et al., 2018; Weerakoon et al., 2015).

Further, in special situations or experimental settings, other alternative diagnostic markers can be used to complete the information about the infection and its pathology. Hematochezia, calprotectin, micro-RNAs, metabolomic markers, apoptotic markers, fibrotization markers, liver transaminases, oxidative stress markers, glutathione, antioxidant enzymes, coagulation markers, and immune cells framework are some to mention. In experimental diagnostics, researches often choose multi-level examination of a certain pathology. For instance, fibrosis-associated genes, proteins, and affected tissue are analyzed (McManus et al., 2018; Weerakoon et al., 2015; Ross et al., 2002).

The following chapter (3) reviews these introduced diagnostic methods in detail in specific challenging settings and applications.

3. DIAGNOSTIC CHALLENGES

Despite decades of advances and encouraging success achieved in the fight against schistosomiasis burden, a few significant challenges remain to overcome (Rollinson et al., 2013).

Several countries showed progress in schistosomiasis elimination while others are not even close to successful control, so the disease persists with high prevalence (Engels et al., 2002; Chitsulo et al., 2000). Tools for sustainable control are currently available, and new ones are being implemented and improved. However, schistosomiasis affects the world´s most vulnerable communities in resource-restricted settings, where is a need for specific and well-adjusted approaches (Ajibola et al., 2018; King, 2010). There is an obvious disparity between sub-Saharan Africa, which has the highest disease burden and the rest of the world. In a lot of these developing countries, schistosomiasis control is almost non-existing. They face other major political, economic, and social challenges and seemingly more dangerous health threats. Such countries are unable to employ successful schistosomiasis control on their own and require international help (Tchuem Tchuenté et al., 2017; Engels et al., 2002).

As we know from previous chapters (1.2, 1.3), published schistosomiasis epidemiological data are often incoherent. National control programs and surveys seem to provide the most reliable information (Engels et al., 2002). However, in these programs, microscopic detection of eggs with all introduced limitations (chapter 2.1), particularly low sensitivity creating a number of false negatives is employed as diagnostics of choice (Weerakoon et al., 2015). On the epidemiological level, every missed infection can restart the transmission hindering the control efforts. Besides disease burden underestimation with all associated consequences, misdiagnoses will obviously affect the patients. Every misdiagnosed individual can suffer from disability and morbidities associated with the disease and develop chronic pathology later the time.

Furthermore, proper implementation of preventive MDAs, the current control strategy of choice, also requires more accurate diagnostics than microscopic detection of eggs, especially in those countries allowing only selective treatment (Ajibola et al., 2018; Bergquist et al., 2017; Weerakoon et al., 2015). The majority of MDA campaigns are targeted through school systems on pupils as they are thought to be most endangered. However, other population subsets such as pre-school children and adults with developed chronic morbidities (e.g., FGS) are also in need of therapy (McManus et al., 2018; Le and Hsieh, 2017). Note to warn again that extensive single-drug MDAs, especially when improperly targeted may seriously affect the *Schistosoma* spp. biology and dynamics with potentially threatening consequences (McManus et al., 2018; Bergquist et al., 2017). With the introduction of more accurate testing methods, control programs can move from MDAs to the more effective “test and treat” scheme (Tchuem Tchuenté et al., 2017).

What can be also improved with adoption of reliable diagnostics is the required switch from current morbidity focused control to more eligible goals such as interruption of transmission, preventing reinfection, and reducing prevalence (Rollinson et al., 2013; Bergquist et al., 2009). This is a long-term mission that requires changes in both methodology and management (Weber et al., 2019; Tchuem Tchuenté et al., 2017).

In effect of successful control and elimination of schistosomiasis, new challenges may arise. These integrated strategies change the disease distribution, reduce the prevalence, number of people at risk, as well as infection intensities. Therefore, more suitable diagnostics, to avoid inadequate assessments is strongly required (McCarthy et al., 2012; Cavalcanti et al., 2012; Engels et al., 2002).

3.1 Acute schistosomiasis

Acute schistosomiasis is introduced above in chapter (1.1) as the first systemic hypersensitive body reaction to the infection. In the symptomatic patients, who are usually travelers from non-endemic countries, it commonly has an unforeseen onset and many non-specific symptoms such as fever, dry cough, headache, diarrhea, malaise, fatigue, urticaria (Jauréguiberry et al., 2010; Ross et al., 2007). Besides these symptoms, eosinophilia, and chest radiography findings of pulmonary infiltrates, bronchial thickening are often observed. Due to this complex symptomatology, acute schistosomiasis is often misdiagnosed and unrecognized by specialists (Ross et al., 2007). In most patients, the syndrome disappears spontaneously after 2 to 10 weeks, while in some others, life-threatening pulmonary, neurological, and cardiac complications requiring the specific diagnosis may occur (Jauréguiberry et al., 2010; Gryseels et al., 2006). Diagnosing and later treating patients with acute schistosomiasis, even the asymptomatic cases, is very important because it can later develop severe pathologies, and even a single untreated individual may represent the potential threat of establishing a transmission site (Cleninx and Van Gompel, 2010; Ross et al., 2007).

Generally used microscopy and serology may fail in diagnosing early schistosomiasis because both oviposition and sometimes even seroconversion may not be present at the time of clinical presentation (Jauréguiberry et al., 2010; Ross et al., 2007). Specific anti-schistosomal antibodies appear approximately after 4 to 7 weeks post-infection as a first detectable diagnostic marker (McManus et al., 2018). Most of the commercial ELISA kits use egg antigens, which are less sensitive for acute syndrome than the use of adult worm or larval antigens may be (Jauréguiberry et al., 2010; Ross et al., 2007). These tests are recommended to be repeated several weeks after schistosomiasis is suspected. Other clinical tests, such as CBC and radiology, can also be performed to confirm the diagnosis (McManus et al., 2018; Lambertucci et al., 1997). History of freshwater exposure in endemic areas followed or not by skin dermatitis, together with marked eosinophilia, is highly suggestive of the acute syndrome and should lead the examiner to the suspicion of waterborne parasitic infection. This diagnostic examination is more suitable for travelers than endemic areas residents (Clerinx and Van Gompel, 2011).

In conclusion, challenging acute schistosomiasis diagnosis in travelers is often not made until years after their return from endemic areas (Corachan, 2002). The diagnosis should be made based on travel history, together with the multiple tests results (Gray et al., 2011). Advanced travel medicine provides better insight to disease progress in non-immune patients and can contribute to the determination of at-risk locations (Corachan, 2002).

3.2 Chronic schistosomiasis

Chronic schistosomiasis is introduced above in the chapter (1.1) as a complex immune system reaction to egg deposition and granulomatous formation in body tissues, usually in endemic countries residents. Patients with the chronic urogenital disease usually have symptoms such as abdominal pain, altered painful urination, and hematuria; manifestations including bladder and ureter fibrosis, kidney damage, genital lesions, vaginal bleeding, prostate damage (Le and Hsieh, 2017; Gray et al., 2011; Gryseels et al., 2006). Patients with the chronic intestinal disease usually have symptoms such as abdominal pain, altered painful defecation, and hematochezia; manifestations including colitis, polyp formation, hepatosplenic inflammation, hepatic fibrosis, portal hypertension, pulmonary hypertension, hepatomegaly, splenomegaly (Olveda et al., 2014; Gray et al., 2011; Gryseels et al., 2006). Associated complications of both chronic diseases include co-infections, infertility, cancer (van Tong et al., 2017; Ross et al., 2002).

The worm burden and egg excretion in these patients are generally low (McManus et al., 2018). Therefore, the direct microscopic detection of eggs in the excreta is not reliable on its own but can be useful in combination with other approaches (Colley et al., 2014; Gryseels et al., 2006). Subclinical patients with morbidities that cannot be detected by microscopy are commonly misdiagnosed (Weerakoon et al., 2015).

Questionnaires, as a first-step approach followed by field-adjusted imaging techniques, showed satisfying results in determining chronic schistosomiasis in such highly endemic areas (Le and Hsieh, 2017). For hepatic disease, the diagnostic gold standard is liver tissue biopsy, which provides satisfying results. Other affected organ biopsies are also common (Weerakoon et al., 2015). However, this approach is invasive, with the risk of the potential development of complications following the procedure. A biopsy examination is a specific method but may lack the required sensitivity (Le and Hsieh, 2017).

Medical imaging is a valuable approach for clinical diagnoses, and accurate pathology assessment of chronic disease (McManus et al., 2018). Ultrasonography is commonly used and can reveal a wide range of symptoms, including some highly specific for schistosomiasis such as periportal fibrosis (Symmer's clay pipe stem fibrosis), liver lesions (Bull's eye lesions), hypertrophy, atrophy, gallbladder wall thickening, nodules. Other imaging methods, CT and MRI, are used less often and show similar findings as US covering also splenomegaly, ascites, accentuation, and portal vein thrombosis. The specific pattern observed by CT in *S. japonicum* infected liver appears like turtleback lesions (Weerakoon et al., 2015; Olveda et al., 2014). In conclusion, medical imaging is a valid tool for diagnosing chronic schistosomiasis in both clinic and field community level (Norseth et al., 2014).

In basic clinical testing, progressive inflammation markers, iron-deficiency anemia, and eosinophilia can be observed (Colley et al., 2014). Non-invasive detection of fibrotic markers is a promising method for chronic schistosomiasis diagnostics in clinics as well as in field-based screening (Weerakoon et al., 2015). Extracellular matrix molecules such as collagen, hyaluronic acid, laminin, and others have the potential to be used as a routine tool for evaluation of schistosome induced fibrosis. However, they require further research and optimization. Other biomarkers like serum antibodies, cytokine levels, parasite antigens, circulating parasite microRNAs, and DNA are currently used mostly for specific clinical cases in specialized laboratories and for research purposes (McManus et al., 2018; Weerakoon et al., 2015; Ross et al., 2002).

The specific end-stage manifestation of chronic schistosomiasis with many associated risks, female genital schistosomiasis (FGS), develops when ectopic egg deposition occurs in the female genital tract (Colley et al., 2014). The complexity of this disease presents a challenge in clinical diagnosis as well as in control programs (Norseth et al., 2014). Direct microscopy is highly insensitive as the eggs retain trapped in the tissue. This pathology can persist without the presence of active infection, making direct parasitological methods insufficient (Colley et al., 2014). Regardless of the limitations mentioned above, a biopsy of genital tissue is a current standard for diagnosing FGS. Colposcopy and US, available also for field applications, are considered to be more secure approaches. In this specific case, direct female genital tissue examination comes across also cultural and ethical obstacles. Diagnosis of male genital schistosomiasis (MGS), less prevalent than FGS, presents with the same limitations. Accurate diagnosis of genital schistosomiasis is complicated and needs to be improved (Le and Hsieh, 2017; Norseth et al., 2014).

Ectopic deposition of eggs during chronic disease may occur in many different organs, including brain, spinal cord, lungs, dermis, spleen, etc. (McManus et al., 2018; Gryseels et al., 2006). In such cases, definitive diagnosis is confirmed with specific methods that are chosen according to the symptoms and manifestation. For example, neuroschistosomiasis is confirmed by neuroimaging and biopsy (Weerakoon et al., 2015), chronic pulmonary schistosomiasis by chest radiography, biopsy, and echocardiography (Bethlem et al., 1997).

An adequate combination of these approaches, together with a comprehensive history, should provide a complete diagnosis of chronic schistosomiasis (Gray et al., 2011).

3.3 Two different worlds

The world of schistosomiasis diagnostics is split into two different worlds (see figure 4) of high-resource unlimited settings with ultra-modern healthcare facilities in developed countries and poor-resource limited settings with minimal healthcare provision in endemic countries (Ajibola et al., 2018).

Figure 4 / Two different worlds of schistosomiasis

The visual representation of significant disparity between two worlds of not only schistosomiasis diagnostics. (A) Well-equipped laboratory with the latest ultra-modern laboratory devices in unlimited settings. Figure adapted from Swiss Tropical and Public Health Institute (Swiss TPH); swisstph.ch/en. (B) Common laboratory settings in the endemic areas with limited resources. Available equipment allows performing only very basic examinations and procedures. Figure adapted from the Schistosomiasis Control Initiative (SCI) organization; schistosomiasiscontrolinitiative.org/tanzania.

Diagnosing schistosomiasis in travelers and migrants in specialized laboratories and hospitals possess different requirements and approaches. The test accuracy is here in the first place. Hight-tech equipment, skilled and experienced laboratory staff, professional physicians, and direct link between many hospitals and research sector allow the use of the most accurate and advanced techniques without basically any great limitation (Clerinx and Van Gompel, 2011). These patients are most likely to present acute schistosomiasis, which manifestation is a lot different from the chronic disease most prevalent in endemic countries (Lingscheid et al., 2017; Corachan, 2002). Accordingly, the diagnosis must be different. The major challenge in such settings appears to be the examiners' awareness of this non-endemic tropical disease and knowledge of its multi-faced nature (Clerinx and Van Gompel, 2011; Corachan, 2002).

The problem is that only a small number of global schistosomiasis cases get to be diagnosed in such settings. Millions of infected people live in developing countries with highly limited resources (King, 2010). Most disease control programs aim at the infections but neglect the complexity of economic, political, and social factors underlying the general state of healthcare (Savioli et al., 2017; Engels et al., 2002). In developing countries, there are a couple of challenges and limitations to the deployment of required diagnostic approaches. In such settings, cost-effectiveness, user-friendliness, and simple logistics are motive factors, often at the expense of the test accuracy (Ajibola et al., 2018). The favorable test for endemic countries satisfies so-called ASSURED criteria, meaning it is Affordable, Sensitive, Specific, User friendly, Rapid, Equipment-free, and Deliverable to those in need (Hinz et al., 2017). Point-of-care, field-adjusted, mobile testing enabling the diagnosis of the remotest, hard to reach communities in poorly serviced locations should be encouraged rather than complicated transportation of patients or samples to distant laboratories (Le and Hsieh, 2017). Advanced and improved methods suitable for diagnostics in resource-limited settings are essential for the control and elimination of schistosomiasis (Ajibola et al., 2018).

The two different worlds disparity is not shown only at the choice of diagnostic standards and the state of healthcare but also at other conditions. For example, in developed countries, access to safe water, proper sanitation and hygiene is a matter of course but not at all in developing countries (King, 2010). In a lot of mostly sub-Saharan countries, schistosomiasis control is rejected and neglected by national authorities who feel that the implementations are unreachable in their settings. That leads to a loss of diagnostic potential in public health care units resulting in the persistence of schistosomiasis (Engels et al., 2002).

3.4 Control programs and epidemiological situation monitoring

Schistosomiasis control programs are done through multiple stages as the epidemiological situation changes. These stages include mapping of prevalence and morbidity to set a baseline of infection status and correctly target MDAs; monitoring of the transmission level and the impact of applied interventions and their gradual reduction; and the final-stage surveillance after successful elimination, with control of possible reemergence of the disease (Rollinson et al., 2013; McCarthy et al., 2012). The success of all these elimination stages depends strongly on the accuracy of the chosen diagnostics. The successful schistosomiasis control program, by its nature, requires diverse diagnostic tools accurate for each stage (see figure 5; Bergquist et al., 2009). For the WHO goal to expand the control and interrupt the transmission with the MDAs as the main tool, repeated screening with reliable methods is necessary (WHO, 2013a). A single general approach cannot accurately reflect the elimination process in all of its stages. In these diagnostic situations, there is a search for a compromise between quantity and quality, research-relevant high-tech approaches are not always applicable for field use (Weerakoon et al., 2015; Cavalcanti et al., 2013).

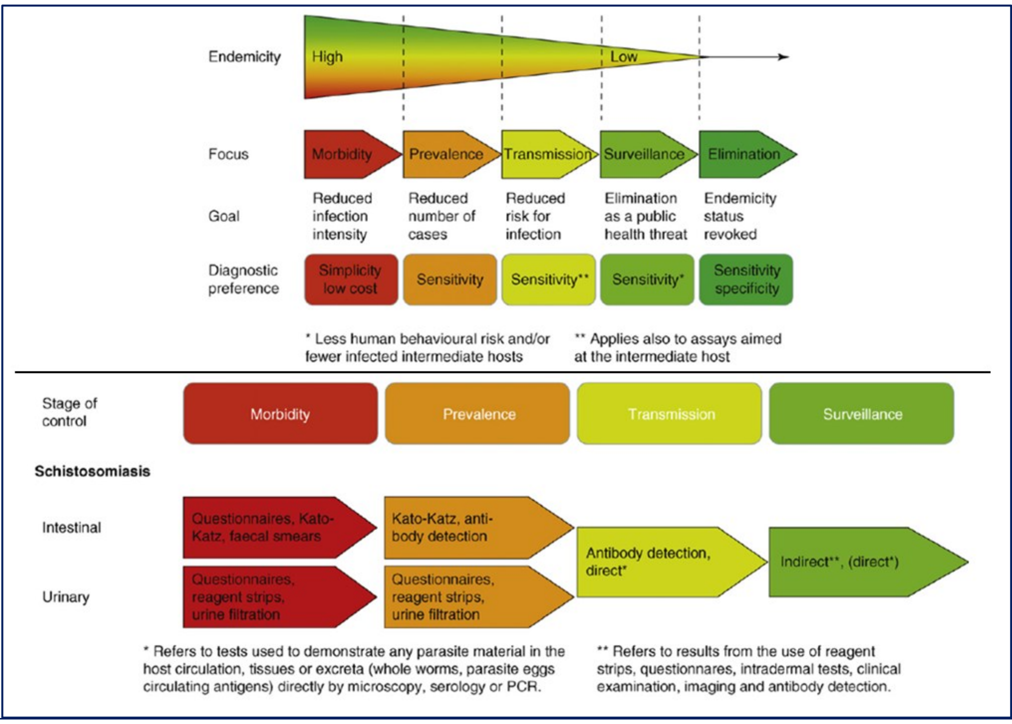

Figure 5 / Suggested choice of diagnostic tools accurately reflecting different stages of disease control
Implementation of control strategies and interventions change the disease endemicity levels and influence its burden. Accordingly, to these changes, the choice of accurate diagnostic tools should be made. This schematic figure shows how reduced endemicity transforms the control focus, goals, and diagnostic requirements. The second part of the scheme represents the suggested use of currently available diagnostics for schistosomiasis in different stages of its control. Figure and information adapted from Bergquist et al., 2009

Any of the mentioned diagnostic methods in chapters (2.1 to 2.5) may be useful for epidemiological studies and control if suitably applied and combined. In the first stages of morbidity and prevalence control, questionnaires especially when combined with urine reagent dipsticks proved to be valuable approaches (Le and Hsieh, 2017; Weerakoon et al., 2015). Standard microscopy is recommended too at these stages. However, unspecific use of this approach may deliver inaccurate results (Bergquist et al., 2009).

Further, in control stages, antibody-based approaches are valuable, and in combination with other tools, are recommended in low-endemicity to confirm and map the interruption of transmission. Antigen-based methods that can be potentially used for these purposes too, still require optimization for field applications (Hinz et al., 2017; Cavalcanti et al., 2013). However, in control of *S. mansoni* and other intestinal schistosomiases, POC-CCA has the potential to become as valuable as urine reagent strips for *S. haematobium* urogenital infections (Le and Hsieh, 2017; Bergquist et al., 2016). The DNA based techniques are slowly becoming more available for field use yet remain 'luxurious' methods. Optimized DNA testing could serve as a relevant tool for field diagnosis and surveillance (Weerakoon et al., 2018).

Fortunately, there is a variety of available or being developed methods that can be optimally implemented in any stage of control programs (Ajibola et al., 2018). These advances with the right management will provide the success of elimination strategies. The use of reliable diagnostics in every stage should be partnered with proper multi-level socioeconomic, environmental, and educational interventions (Tchuem Tchuenté et al., 2017).

3.5 Follow-up care, reinfections, and light infections

How to accurately measure anti-schistosomal treatment response and efficacy is still a matter of debate (Weerakoon et al., 2015).

For large-scale applications traditionally direct microscopic diagnosis followed by quantitative evaluation of changes in infection intensities before and after treatment are used (Montresor, 2011). These methods provides cure rate (CR), which represents the percentage of before treatment positive patients with negative excreta sample after the intervention and egg reduction rate (ERR), which represents the percentage of decrease in a number of excreted eggs (Olliaro et al., 2015). There is an available guideline for ERR implementation, which is considered to be more suitable than CR, as the standard approach for follow-up surveys measuring the MDAs outcomes and morbidity changes (WHO, 2013a). However, as these methods are based on microscopic detection, they have limitations in providing conclusive results, which may also differ from area to area due to the different baseline setting (Plliaro et al., 2015; Montresor, 2011). Urine reagent dipsticks for diagnosing urogenital schistosomiasis also indirectly correlate with the infection intensity so can be used as well for this purpose (Le and Hsieh, 2017). Stete et al., 2012, recommend measuring ERR from day 14 after-treatment, CR and urine dipsticks at week six after treatment for accurate monitoring of PZQ antischistosomal effect against *S. haematobium* in endemic regions.

A much less common approach for follow-up surveys is antigen detection, which has the exact characteristics required in therapy evaluation (Hinz et al., 2017). Imaging techniques are not directly specific in evaluating the treatment effect but can be used as a supportive method (Olveda et al., 2014). DNA-based detection can also evaluate after-treatment conditions (Weerakoon et al., 2018).

Follow-up care of imported schistosomiasis includes monitoring of active disease parameters such as clinical symptoms, eosinophilia, and the presence of schistosome eggs in the excreta (Praticò et al., 2014). Schistosomiasis parameters decrease in treated patients gradually. Eggs in excreta can still be detected several weeks after successful treatment (Stete et al., 2012). Medical imaging is useful to assess the reversal and improvement of schistosome induced chronic pathologies after the treatment (Olveda et al., 2014). Detection of antibodies does not reflect the treatment response accurately. Antigen and DNA detection can be used as supportive methods in follow-up care. In patients with recurring symptoms, the full overall diagnostic examination should be repeated (McManus et al., 2018; Weerakoon et al., 2015; Gryseels et al., 2006). At least two years of general follow-up care after initial treatment administration are recommended (Praticò et al., 2014).

Schistosomiasis reinfections and disease reemergence diagnostics require approaches with similar features as follow-up care. Thus the same or similar methods are used for this purpose (Mutsaka-Makuvaza et al., 2018). Reinfections occurring within a few days or weeks after treatment administration are undetectable with currently available diagnostic methods and might be reported as failed treatment (Stete et al., 2012). Studies of reinfections after confirmed successful treatment use direct microscopy with ERR and CR quantification, urine reagent strips, and questionnaires (Woldegerima et al., 2019; Mutsaka-Makuvaza et al., 2018). Other suitable approaches, such as antigen and DNA detection also have the capacity to diagnose reinfection (McManus et al., 2018; Weerakoon et al., 2018). Detected reinfection cases usually are low-intensity infections (Woldegerima et al., 2019). Problematic diagnostic assessment of these studies requiring repeated sample collection and examination over a sustained period of time remains to be a challenge for control and elimination programs (Ajibola et al., 2018).

Currently, increasing prevalence of light infections as a result of MDAs in endemic areas brings out the need for highly sensitive approaches. As mentioned before, direct microscopy is insensitive in diagnosing light infections, but in combination with concentration methods, it can be useful for this purpose (Utzinger et al., 2015; McCarthy et al., 2012). In low endemicity areas, the suggested diagnostic scheme is to use antibody-based techniques in initial population screening. Positive serology cases then should be tested either with antigen or direct egg detection to confirm the diagnosis. After treating these positive patients, the surveillance is suggested to be done with rapid tests or DNA-based techniques (Cavalcanti et al., 2012; Bergquist et al., 2009). Clinical diagnoses of light infections in travelers are commonly confirmed by antibody detection, along with direct parasitological methods and basic laboratory testing such as CBC (Jauréguiberry, 2010).

3.6 Experimental infections

Great contributors to a better general understanding of schistosomiasis disease are experimental infections. For research studies of basic biology, immunology, pathology, pathogenesis, as well as for product development, diverse experimental models are used (Cheever et al., 2002). The majority of these models are animals, especially mice in which the disease responds differently, and the standardized human diagnostic methods might not always be suitable to use (Cheever et al., 2000).

Diagnostics applications used in experimental research are very specific. Methodological requirements differ according to the study's purpose. It is often required to accurately assess every change in disease progress, to measure all the infection aspects, to conduct real-time observations of various characteristics, to evaluate efficiency and mechanisms of tested drug or other therapy, and to compare the data to control standard (Maezawa et al., 2018; Cheever et al., 2002). Note to say, the present review is focused on *in vivo* testing. Experimental studies can be evaluated at many levels by many diverse techniques.

Besides general health monitoring of the models, popular methods in these studies are the microscopic examination of eggs in excreta, together with egg burdens in affected tissues (e.g., liver), and oogram profile (= the state of egg maturation). This standardized schistosomiasis characteristic widely used in diagnostics displays the chronic aspect of the infection and indirectly also the worms' physical state.

Furthermore, counts, viability, egg production, pairing rate, and various morphological and developmental alterations of adult worms recovered from the experimental models by perfusion are observed under a microscope, often using high-resolution techniques (Ellakany et al., 2019). Gross tissue pathology examination after parasitological dissection followed with tissue-specific microscopy, histopathology, and immunohistochemistry, together with the analysis of granulomas load, size, composition, and tissue weight index, are valuable approaches (El-Lakkany et al., 2012). Specific proteins and markers, for instance, fibrotization and stress markers, liver damage enzymes detection are used to obtain complex data of the disease state. For the evaluation of immunological state, cytokine, antibody, and antigen levels are measured by ELISA and other immunological assays (Aly and Mantawy, 2012). Nucleic acids-based techniques to detect parasites specific RNA and DNA or, for example, also human fibrosis-related genes expression are also measured in specific studies (El-Shennawy et al., 2015). Imaging techniques adapted for experimental models provide non-invasive insight on various disease characteristics (Maezawa et al., 2018). An optimal combination of multiple methods is chosen by the majority of authors and provides the best representative and comparable results.

Experimental studies greatly contribute to the general understanding of the disease as well as to the development, implementation, optimization, and standardization of novel diagnostics and therapy of schistosomiasis (Weber et al., 2019).

4. MOVING TOWARDS ELIMINATION OF SCHISTOSOMIASIS

When a proper diagnostic assessment is provided, what else is required and suggested to achieve success in fight with schistosomiasis?

The road towards the elimination of this multifaced disease with complex characteristics and intricate burden requires renewed global commitment and information (McManus et al., 2018; Tchuem Tchuenté et al., 2017). Successful control programs depend on local political, economic, social, and environmental factors of every endemic country influencing the application, adoption, and response to the implemented strategies (Weber et al., 2019; Rollinson et al., 2013). This should be taken into consideration while designing schistosomiasis control programs. Integrated interventions should include latest advances from research and development, reliable diagnostics, preventive chemotherapy with adequate targeting, improved water and sanitation provision, promoted awareness, hygiene education and behavioral changes, organized snail and environmental monitoring, plus sustainable multi-level cooperation and partnerships between all involved subjects (Grimes et al., 2015; Rollinson et al., 2013).

Based on the available knowledge, experience, and novel expertise, the strategic research agenda should reconsider the use of current control standards and validate the new promising ones. Advances from fields such as biology, ecology, medicine, epidemiology, economy, and others can help to identify the challenges and introduce optimized methodologies and guidelines to the national control programs (McManus et al., 2018; Colle et al., 2014). This connection between laboratories and field and proper implementation of research findings into commercial use is the backbone of schistosomiasis elimination success. The research field should be well involved in these strategies (Weber et al., 2019).

In the following paragraph some research areas important in moving towards schistosomiasis elimination are listed. ✔ Accurate disease diagnostics and assessment with improved, standardized, rapid, field-applicable tools available at POC (Weerakoon et al., 2015). ✔Renewed PZQ formulation and the development of new anti-schistosomal therapeutics and vaccines (Rollinson et al., 2013). ✔Better understanding of clinical burden, pathologies, mortality, neglected morbidity aspects, and their consequences. ✔Studies and evaluation of zoonotic cycles and animal reservoirs together with better treatment methods and formulations for infected animals. ✔Genetic studies of parasite-host interactions and genomic characterization of parasites and their intermediate hosts (McManus et al., 2018). ✔Eligible determination of environmental changes, global migration trends, and water bodies engineering impacts. ✔Mapping of disease hot-spots and high-risk areas disease dynamics patterns (Steinmann et al., 2006; Chandiwana and Woolhouse, 1991). ✔Evaluation of the socio-economic impact of schistosomiasis (King, 2010). ✔Implementation of mathematical modeling and measures in the disease studies (McManus et al., 2018).

As pointed out before, the major contributor to disease transmission is limited access to safe water and poor sanitation. Preventing the water from excreta contamination is the key intervention not only for schistosomiasis but also for other water-related diseases. Socioeconomic improvements in so-called WASH components – water, sanitation, and hygiene together with intensive health education and behavioral changes promotion are necessary to stop the contamination and disease transmission (Campbell et al., 2018; Grimes et al., 2015).

Another important part of elimination strategies is the environmental control of intermediate hosts (Weerakoon et al., 2015). The application of molluscicides can interrupt the transmission. However, it raises the expenses of the control initiative. Such an intervention has to be used with care as it could influence the whole ecosystem. More ecological-friendly alternatives to the chemicals, for example, the introduction of molluscivorous fish into the targeted sites, can be beneficial (Stothard et al., 2017).

The global community is determined to eliminate the schistosomiasis (Weber et al., 2019; Savioli et al., 2017; WHO, 2013b). For priority areas of global schistosomiasis alliance (GSA; see figure 6). Eventually, it is cross-sector partnerships and collaborative agreements that will create a sustainable framework to fight schistosomiasis as well as other NTDs (Weber et al., 2019). For instance, the control of onchocerciasis or trachoma benefits from organizations collaborations and is further than current schistosomiasis control (Savioli et al., 2017). This cooperative approach and active communication between local authorities, health facilities, governments, non-governmental organizations, academic and private sectors, and many others are essential (Weber et al., 2019).

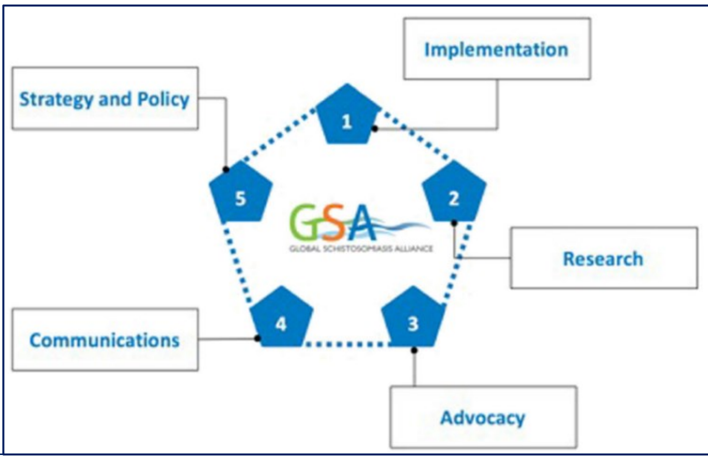

Figure 6 / Priority areas of global schistosomiasis alliance (GSA)

This scheme represents five key interconnected themes in GSA's focus, pivotal in the aim of schistosomiasis elimination. (1) Implementation refers to the provision of improved hygiene and education components, preventive chemotherapy as well as other operational support. (2) Promising research areas should be encouraged, and novel advances introduced and optimized. (3) Coordinated advocacy between endemic and non-endemic countries and mobilization of regional facilities to boost the improvement and raise the awareness. (4) A variety of communication tools is required to deliver all the information in between all the associates. (5) After properly established schistosomiasis elimination mission efficient strategy and policy is needed to ensure the right cooperation and allocation. Figure and information adapted from Savioli et al., 2017

As mentioned before, some endemic countries lack resources and capacity to run a successful control program with full chemotherapy coverage and accurate assessment (Engels et al., 2002). Allocations of available funds, charity, and drug donations could be communicated through these partnerships. It is beneficial to have an international platform for standardization of control tools such as diagnostics, therapy, water and sanitation plants, education, as well as for incentivization of the private sector into research and development for NTDs (Weber et al., 2019; Tchuem Tchuenté et al., 2017; Savioli et al., 2017). In the best scenario, these global alliances should help the most vulnerable countries to implement disease control as the internal part of national healthcare and overcome the difficulties to become self-sustainable with no need for international help (Tchuem Tchuenté et al., 2017; Engels et al., 2002).

With all these improvements, every day, we are closer to the schistosomiasis elimination.

CONCLUSION

A lot of information about schistosomes and schistosomiasis was gathered during more than 150 years since these infectious parasites responsible for the disease were described. Control programs with large-scale chemotherapy support has been more or less successfully implemented for decades. However, the global burden of schistosomiasis is still disturbingly high. Published epidemiological data are often incoherent and disconnected from reality. Mortality levels and overall impact of this disease are generally underestimated. The use of unsuitable and inaccurate diagnostic standards for various settings is partially responsible for these issues.

The most appropriate application of diagnostic methods should be based on the combination of their key values such as accuracy, sensitivity, specificity, rapidity, cost, and convenience that adequately cover the situation. In non-endemic developed countries, sensitive and specific tests are preferred regardless of the price. On the other hand, in highly endemic countries of developing worlds, cheap tests are favored even though those currently available express low sensitivity.

In conclusion, the collection of available laboratory-based techniques for diagnosing schistosomiasis is broad, and new methodologies are being introduced in most recent publications. Microscopy-based methods are useful in highly endemic areas and individual diagnosis. Antibody-based assays are relevant in diagnosing travelers, in post-elimination settings and low-endemicity areas. Antigen and parasite DNA detection is valuable tool for research purposes and with further optimization also promising for field use. Along with laboratory methods, there are also relevant and valuable non-laboratory techniques such as questionnaires and medical imaging. There is a potential to adopt a variety of methods suitable for use in endemic areas along with the direct microscopy. There is an operational space to make the right choices. The choice of an adequate diagnostic method is essential for making the correct schistosomiasis diagnosis and in the bigger picture for elimination this debilitating disease.

List of figures

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